Armed Forces Radiobiology Research Institute

Retrospective Reconstruction of Radiation Doses of Chernobyl Liquidators by Electron Paramagnetic Resonance

Scientific Center of Radiation Medicine
Academy of Medical Sciences, Ukraine

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Preface

On April 26, 1986, Reactor #4 at the Chernobyl Nuclear Power Plant near Pripyat, Ukraine, exploded, releasing millions of curies of radioactive materials into the environment. The reaction was swift, with firefighters and medics being mobilized within hours of the accident. After initial care of the injured and instituting measures to prevent further exposure of the general population in the vicinity, attention was turned to cleanup of the damaged reactor and the radioactive debris. Hundreds of thousands of workers (called "liquidators") were employed in the cleanup. Authorities were aware of the risks of immediate and long-term health effects to these people and took measures to limit the dose received.

With the passage of time, the liquidators have developed leukemia, solid tissue neoplasms, cardiovascular disease, and other illnesses. The question of what relationship these illnesses, which also occur in unexposed populations, bear to the radiation exposure received at Chernobyl naturally arises. This question is vitally important, not only for compensation purposes, but also for advancing our knowledge of the effects of protracted radiation exposure on human health and for setting or reevaluating safety standards. But the critical first step in finding the answer is accurately ascertaining what dose was actually received. Physical dosimeters were not always used, and were not always used reliably, during the several operations involved in the Chernobyl cleanup. It is necessary to employ accurate, reliable biological indicators of radiation effects to reconstruct exposure received.

The implications of using electron paramagnetic resonance (EPR) analysis as one such state-of-the-art technique in performing dose reconstruction clearly go beyond Chernobyl. There are other areas of the world with widespread environmental contamination at dose levels sufficient to cause adverse health effects, such as along the Techa River in the Southern Urals region of Russia and in the areas surrounding the former nuclear weapons test site near Semipalatinsk, Kazakstan. There have also been accidents involving small numbers

of individuals where the actual dose received is, for one reason or another, not accurately known.

Because of the importance of a means of accurately and precisely estimating cumulative radiation exposure for epidemiologic studies, the Armed Forces Radiobiology Research Institute (AFRRI) elected to fund this study. The authors are highly competent investigators who also have connections with similarly skilled independent scientists who could refine their techniques and improve their results. In addition, they have access to the data from the Chernobyl liquidators accessible to followup, most of whom are now in Ukraine. Although the scope of this study was limited, its results should provide a significant step in improving the utility of EPR in dose reconstruction as well as in getting a clearer picture of the magnitude of the radiation exposure actually received at the world's most tragic reactor accident.

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Contents

Abstract

Accurate, reliable dose reconstruction is a critical component in the epidemiological followup of liquidators. Dosimetry of teeth by electron paramagnetic resonance (EPR) is a state-of-the-art laboratory technique that is key to this effort. The Scientific Center of Radiation Medicine (SCRM) has developed and refined this technique in order to meet the practical demands of large-scale epidemiologic followup of the Chernobyl liquidators. Independent analysis using similar technology was performed by investigators at the University of Utah and showed good correlation with the SCRM results. The lower limit of detection for reliable dose reconstruction was 100 mGy. Techniques were applied to samples from approximately 135 liquidators involved in cleanup activities within the first 2 years after the Chernobyl accident in 1986. Mean dose was 287 mGy, geometric mean was 205 mGy, and median dose value was 200 mGy. The reconstructed dose values range from 30 to 2220 mGy. Correlation of results between the two institutions was generally within 17%. This report also addresses some confounding factors (previous medical x-ray exposures, ultraviolet light effects on anterior teeth, nonlinearity of dose response curves below 100 mGy) and how to deal with them.

Key words: dosimetry, retrospective, EPR, technique, doses, liquidators, Chernobyl

Introduction

Electron paramagnetic resonance (EPR) dosimetry using teeth is generally accepted as a highly attractive method for reconstructing individual radiation doses long after exposure [1]. However, until recently, EPR dosimetry was generally considered a unique state-of-the-art laboratory procedure unsuitable for practical dose reconstruction. Moreover, the accuracy and consistency of results produced by this technique were not proven.

A tool for dose reconstruction is acutely needed, particularly in the Chernobyl situation. Although these dose records are incomplete and not all liquidators know their doses, an official dose record is included in most of the identification forms of cleanup workers. The objective of the effort funded by DNA (contract number DNA001-95-C-0017) was to develop reliable retrospective estimates of radiation doses received by the Chernobyl liquidators. This goal was approached in three stages.

First, the Scientific Center of Radiation Medicine (SCRM) developed the EPR dosimetry technique to provide reliable and efficient reconstruction of doses. Each of the basic steps of EPR-dosimetric methodology was subjected to rigorous analysis and optimization. Methodological research and development of the technique are far beyond the scope of this contract, but the technique has been explicitly presented elsewhere [2,3] and is widely accepted worldwide. Figure 1 illustrates that every step incorporated a number of innovations and specific features. This improved version of EPR dosimetric technique was employed for both routine dose reconstruction and interlaboratory cross-calibration.

Second, a sophisticated cross-calibration effort was undertaken in order to assure the quality of results. This effort included a series of internal tests as well as intercomparisons with an experienced counterpart in the USA.

Third, after completion of the above tasks, routine reconstruction of doses to liquidators began. The doses to 135 individuals were assessed in accordance with the technique designed in Task One. These results were entered into a database and are available for researchers.

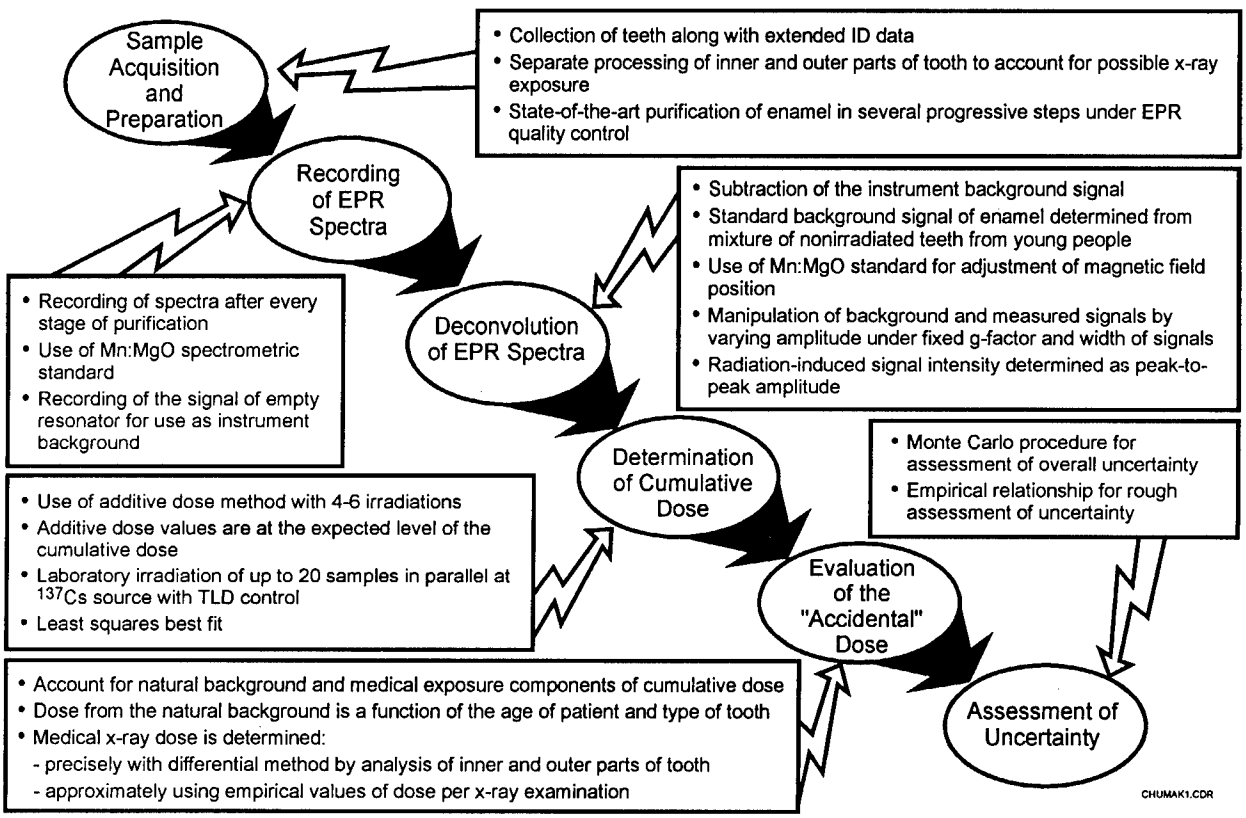

Figure 1. Principal steps of EPR dosimetry and innovative features of the SCRM version of the EPR technique

Task 1
Development of a Routine High-Performance EPR-Dosimetric Technique

Extensive scientific and technological investigations were conducted in order to make EPR dosimetry usable as a routine tool for dose reconstruction. Special attention was paid to ensuring reliable results. With respect to the demands of epidemiological followup, optimal EPR technique must meet the following requirements:

- Sensitivity of the technique and accuracy of the results must be adequate to meet the practical needs of post-Chernobyl followup.
- The results produced by the technique must be consistent with other (independent) dosimetric methods and internal standards.
- The technique must be reproducible at different times and in other laboratories.
- Performance of the technique must be high enough to meet practical demands.

EPR Measurements, Spectra Analysis, and Dose Reconstruction

A brief profile of the EPR measurement procedure is presented in table 1. The use of the Mn:MgO spectrometric standard allows for objective control of stability of the system and ease in accounting for possible drifts and deviations. The standard is used for calibration of the instrument in terms of both sensitivity and g factor. The empty resonator signal is recorded daily and subtracted as instrument background noise in each series of measurements.

Table 1. Brief profile of the EPR measurement procedure used at SCRM

Feature	Characteristics
1. Instrument	BRUKER ECS-106
2. Laboratory irradiator	^{137}Cs
3. Buildup for secondary electrons	+
4. Number of additional irradiations	5
5. Dose increment/cumulative dose values for calibration curve	$\Delta D = 174$ for $D_x < 500$ mGy $\Delta D = 348$ for $D_x \geq 500$ mGy
6. Method of best fit of the calibration curve	Linear regression
7. Sample preparation	+
8. Parameters of EPR registration:	
Microwave frequency	9.81 GHz
Microwave power	10 mW
Center field	348 mT
Sweep width	10.0 mT
Modulation frequency	100 kHz
Modulation amplitude	0.23 mT
Conversion time	164 ms
Time constant	328 ms
Number of scans	15
Measurement time	60 and 45 minutes See item 5
9. Processing of measured spectra and detection of radiation-induced (RI) signals	Subtraction of standard background signal by selectively varying its intensity and fixing g factor and width
10. Type of standard background signal	Spectrum of the mixture of nonirradiated teeth from students 18-25 years old
11. Use of presumably nonirradiated samples as reference	—
12. Approach to the error propagation	For doses less than 500 mGy, the error is determined by uncontrolled impurities of enamel with maximum intensity equivalent to RI signal from 50 to 80 mGy dose; for higher doses, error is determined by statistical error of RI signal determination.

The procedure for mathematical processing was based on investigation of the principal background signal variability. We discovered that background signals of different teeth differ only in terms of intensity, while g factors and width of lines for background signals are constant for all teeth. Therefore, we use a standard background signal, which was obtained from a mixture of several dozen nonirradiated teeth that were extracted from young people (18–25 years old), in order to minimize the natural background dose.

The optimal procedure for subtraction of the principal background signal using the BRUKER ECS-106 or similar instrument is as follows:

1. The standard background spectrum is shifted by the constant magnetic field relative to the sample spectrum, using an Mn:MgO marker until the g factors of both spectra coincide.

2. The amplitude of the standard background spectrum is adjusted to coincide with the sample spectrum.

3. The two spectra are subtracted while keeping the Mn:MgO signal constant.

4. The resultant subtracted g factors of the maximum and minimum components of the suspected radiation-induced (RI) signal are compared to the positions of the relevant points determined for high-dose signals.

5. If points coincide, the intensity of the original radiation signal is measured and the value is used as the first experimental point in the individual calibration curve. If no coincidence occurs, no confident dose reconstruction is possible, and the tooth is considered to be exposed to a dose of less than 0.1 Gy.

This rather conservative approach to spectrum interpretation ensures against the measurement of artifacts and misleading readouts.

To account for individual radiosensitivity of teeth, an internal standard was used. Each sample was exposed to additional doses under controlled laboratory conditions. The ^{137}Cs secondary standard irradiator, calibrated in terms of absorbed dose in air using an 8 mm plastic screen for buildup of secondary electrons, was used for this purpose. The additional doses and the results of subsequent measurement produce the calibration curve of the individual tooth. The intersection of this curve with the abscissa corresponds to the amount of exposure.

Sample Collection

The following samples were collected for use in both practical dose reconstruction and interlaboratory cross-calibration:

- 69 teeth from liquidators (40 were multiple teeth from 15 individuals)
- 19 teeth from unexposed young people in Ukraine
- 6 teeth from unexposed people in the United States

Teeth from cleanup workers were collected in the course of routine dental treatment in the Liquidators' Clinic in Kiev, which is dedicated to the medical and dental care of liquidators. All teeth were extracted for clinical reasons only; none was collected solely for dosimetric purposes. The extraction procedure was performed by a skilled dental surgeon who had been appropriately instructed. Every sample was accompanied by a Tooth ID Form (see appendix), which contains ID information on the patient, a history of occupational and medical exposure, activities and length of stay in the restricted zone, and the officially recorded dose value. After extraction, the teeth were washed with water and dried at room temperature. Each sample was transferred to our laboratory in a clearly labeled, individual container.

Sample Preparation

A new, elaborate method of sample preparation was one of the most efficient innovations introduced into the EPR-dosimetric technique. Application of this procedure leads to the substantial reduction of background EPR signals that normally are superimposed on the radiation-induced signal in tooth enamel, making detection of doses below 0.5 Gy difficult.

When samples arrive at the laboratory, they are registered in the log and computer database. Then sample preparation begins. Since the properties of the original material vary, the degree of purification needed to obtain an optimal specimen may vary. Accordingly, purification is normally performed in several progressive steps.

1. Removal of the tooth root.
2. Splitting of the tooth into its inner and outer parts with a diamond saw (this step is taken in order to take possible medical x-ray exposures into account).
3. Fragmentation of the tooth into 1- to 2-mm particles.

4. Chemical treatment of the tooth with KOH alkaline in an ultrasonic bath for 4 to 7 days to remove dentine and organic components of tooth enamel.

5. Removal of the remainder of the dentine (especially in the tooth parts with the highest curvature) with a hard-alloy dental drill.

6. Crushing of samples to 0.1 to 0.25 mm-sized grains.

7. Additional purification of tooth enamel using a heavy liquid, sodium polytungstate, with a specific weight of 2.92–2.94 g/cm^3.

8. Several washings of samples with distilled water; the last washing takes several hours under ultrasonic processing.

9. If needed, visual control of samples using a binocular microscope to remove nonenamel inclusions.

The failure of any given procedure leads to application of further treatment. This algorithm is graphically presented in figure 2.

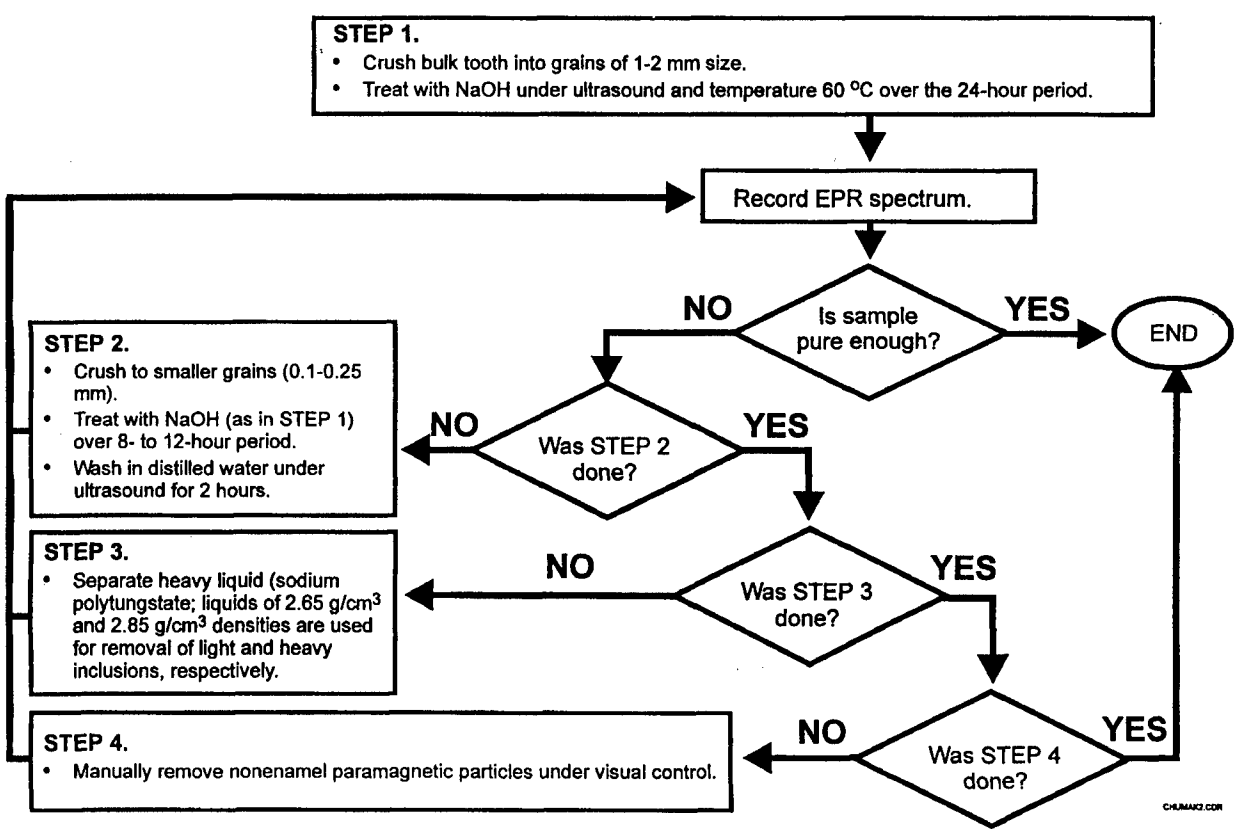

Figure 2. Flow chart of the sample-preparation process

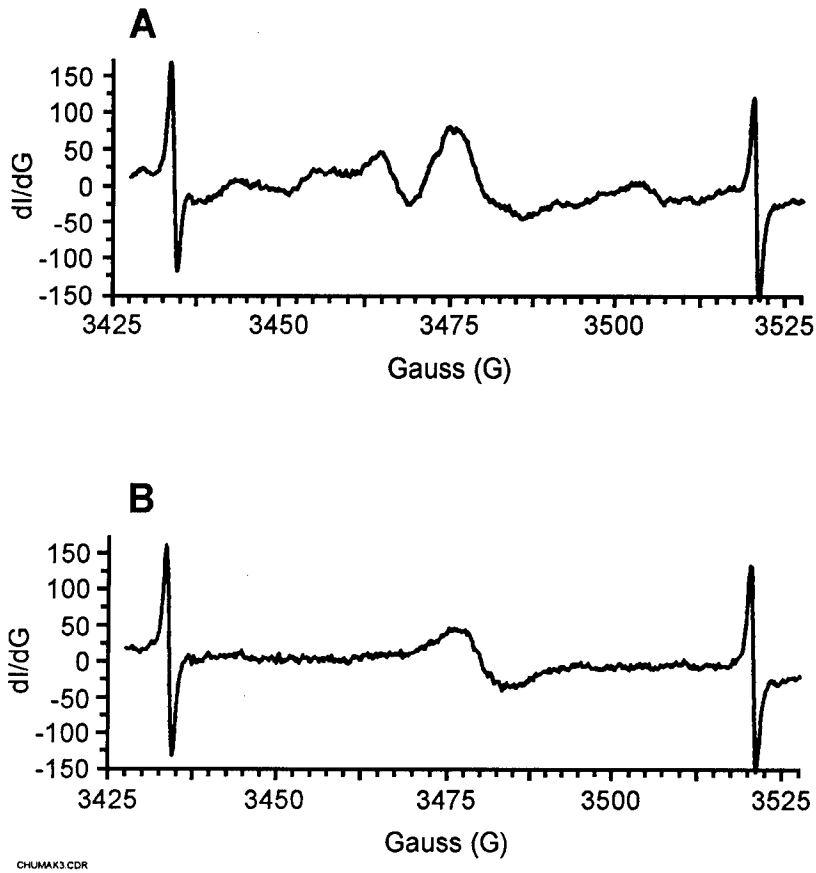

Figure 3. Effect of sample purification. EPR spectra (A) before and (B) after purification of the low-dose sample

Figure 3 shows the EPR spectra of the same low-dose sample recorded before and after application of the purification procedure. Clearly, the spectrum was dramatically improved, making dose reconstruction with the sample possible.

Another important issue is evaluating the contribution of medical x rays to the measured cumulative dose. Oversensitivity of tooth material to low-energy photons is well known, causing serious difficulties in determining Chernobyl-related doses. As much as a 7 to 1 difference in deposited dose in enamel versus that in soft tissue represents a threat to the utility of EPR as a dosimeter. The results of a study of this problem are presented elsewhere [4], giving a clue to a solution to this potentially severe problem.

Task 2
Quality Assurance Program

Dr. Ed Haskell of the Division of Radiobiology, College of Medicine, University of Utah (UU), agreed to take part in the cross-calibration studies. These were performed in three stages, namely:

Stage 1: Intercalibration using samples of tooth enamel with uniform properties that were exposed to known radiation doses under laboratory conditions

Stage 2: Intercalibration using whole teeth exposed *in vitro*

Stage 3: Intercomparison using liquidator's teeth accidentally exposed *in vivo*

Each stage was designed to more closely approximate reality. Thus, the first stage dealt with rather ideal samples while the third intercomparison involved full-scale dose reconstruction using teeth specifically from exposed individuals. Interpretation of the results became increasingly difficult with each stage, as the number of uncertainty factors increased. However, the overall results appear to have engendered confidence in the adequacy of the dose assessments by EPR dosimetry with teeth.

Stage 1. Intercalibration With Homogenized Samples

For the first stage, the simplest intercalibration was performed with unrealistic but extremely uniform samples. This stage could be considered as a check for the reproducibility of the technique for practical dose reconstruction. The significant advantage of the intercomparison design was the possibility of objectively judging the results—no uncer-

tainty factors could lead to a fuzzy interpretation of the dose determinations.

Intercomparison Design

The main purpose of this intercalibration[1] was to test the results produced by different techniques using samples with well-known and uniform properties, thus allowing for objective evaluation of results. Since sample preparation techniques in different laboratories vary significantly, only the minimum necessary treatment was applied to tooth samples.

A large number of nonirradiated human teeth with minimum background doses were treated mechanically in order to extract tooth enamel in the form of 0.1- to 0.25-mm grains. These were mixed together to form homogeneous material. This material was then divided into 100 mg portions and forwarded for irradiation to the IAEA Laboratory in Siebersdorf, Austria. The samples were irradiated with dose levels of about 100, 250, 500, and 1000 mGy. Those receiving doses up to the 500 mGy level were irradiated with a ^{137}Cs source at a dose rate of 800 Gy/min. Those at the 1000 mGy level were exposed to a ^{60}Co source at a dose rate of 200 mGy/min. Sets of samples with five different dose levels (unknown to the participants) were shipped to SCRM and UU. The participants were invited to use their own EPR dosimetric routines (including chemical treatment of samples, EPR measurement, and interpretation of spectra) to determine the exposures received by the samples.

Methods and Results

The intercomparison revealed significant variations in the experimental techniques used for dose reconstruction with EPR of tooth enamel. The SCRM technique is shown in table 1. The major differences in the UU technique were as follows.

- The background signal was not subtracted. The intensity of the RI signal was determined as the difference of intensity at the points corresponding to the g factor of the first maximum and first minimum of the RI signal.

- The samples were measured 10 times each, shaking the tube after every measurement.

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[1] This cross-calibration was performed as part of the First International Intercalibration of EPR dosimetry with teeth, which was sponsored in part by CEC contract COSU-CT93-0051.

- Laboratory irradiation was performed for those samples that demonstrated the most significant variations. For these samples (8 of 14), the value of the initial RI signal was determined as the intersection of the calibration curve with the ordinate axis. For the remainder, the initial intensity of the RI signal was assessed as the mean of 10 measurements. At this point, three samples with minimum RI signal values were considered as empirical zero. Doses of other samples were determined by subtracting the average intensity of three low-dose signals from the RI signal intensity. The results were divided by the value of the average calibration factor derived from the analyses of eight samples.

- The residual signal, which corresponded to the average dose of three nonirradiated samples, was estimated to be 68 mGy.

The results of the intercalibration are presented in figure 4 and table 2. Clearly, the precision of dose determination depends greatly on the dose value and the technique used for reconstruction. The results obtained at SCRM demonstrated an excellent agreement with the preset dose values [5]. On the other hand, the UU technique showed variability over the whole range of doses, from 0 to 1000 mGy.

These results were discussed in May 1995 during the 4th International Symposium on EPR Dosimetry and Applications. The constructive discussion of the peculiarities, possible advantages, and shortcomings of the techniques used at SCRM and UU led to the harmonization of certain approaches used in the two laboratories for subsequent stages of the cross-calibration. The UU approach was changed to conform more closely with some of the particular procedures under the SCRM approach.

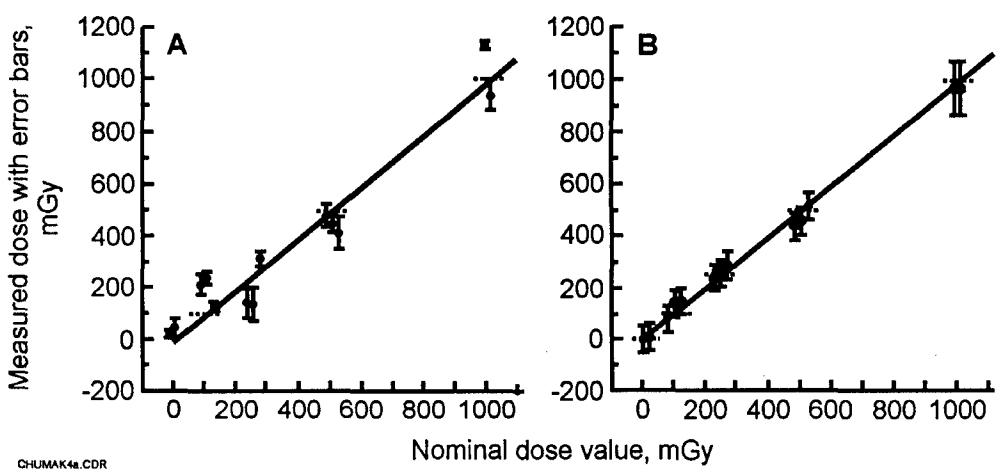

Figure 4. Results of intercalibration with homogenized samples. Measured doses versus nominal dose values. (A) UU, (B) SCRM

Table 2. Results of intercalibration using homogenized samples irradiated in vitro

Participant	Sample number	Measured dose value, mGy	Nominal dose value, mGy
University of Utah	87	20.7±17	0
	88	49.8±28	0
	90	-93±9	0
	21	132±15	100
	4	209±39	100
	6	237±26	100
	35	137±58	250
	43	138±68	250
	58	312±31	250
	75	413±67	500
	66	448±34	500
	56	479±49	500
	106	940±60	1000
	100	1128±21	1000
SCRM	80	0±50	0
	81	10±50	0
	8	80±50	100
	16	140±50	100
	23	150±50	100
	36	240±50	250
	37	260±50	250
	42	290±50	250
	60	440±50	500
	68	460±50	500
	69	520±50	500
	76	970±100	1000
	108	970±100	1000

Stage 2. Intercalibration With Whole Teeth Irradiated Under Laboratory Conditions

The main purpose of stage 2 was to address the possible influence of sample preparation procedures on the results obtained using different techniques.

Intercomparison Design

The sample set consisted of six teeth, collected by a local dentist in the United States. The prior dose of x rays was unknown. Each tooth was split in two at the plane perpendicular to the jaw contour. One half of each tooth was irradiated using a ^{60}Co source with a dose rate of about 10 Gy/h. Measurements were considered to be dependent only upon the total dose, not the dose rate. Three dose levels between 100 and 500 mGy were used; dose was unknown to the participants. One half of each tooth remained unirradiated in order to provide a postmeasurement check for detectable dose due to dental x rays as well as to provide a check should anomalies appear in the spectra or the results.

The participants knew that the pairs of teeth numbered 1 and 4, 2 and 5, and 3 and 6 were irradiated with equal doses in order to provide direct comparison of the results with the equivalent laboratory-added dose.[2] Teeth 4, 5, and 6 were shipped to the SCRM; the others remained at UU. Shipping was done by express mail specially labeled to avoid x-ray inspection and thus minimize transportation dose. Participants were instructed to apply their customary EPR dosimetric technique.

Methods and Results

The samples of tooth enamel were prepared as described in Task 1 above. The tooth size was sufficient to perform separate analyses of the outer and inner parts in order to control, and if possible, account for unreported x-ray examinations.

The EPR spectrum of tooth 6 differed significantly from the expected shape of the signal of irradiated enamel. Chemical processing with NaOH alkali (8 hours in an ultrasonic bath at 60 °C) was used to purify the enamel (see figure 2). Although the subsequent EPR analysis revealed a noticeable improvement, the shape of the signal was still distorted. The specimen was examined under the microscope, and one paramagnetic nonenamel particle was located and removed. That significantly improved the signal, making the sample appropriate for dose reconstruction.

A special effort was made to control for possible x-ray exposure prior to intercalibration. Although the results of this examination do not allow for the reliable quantitative assessment of lifetime dose, there are strong indications of both a dose gradient (i.e., the surface of the tooth nearest the x-ray source receives a higher dose than the surface

[2] The total dose to be measured consisted of two principal components: unknown lifetime dose of the tooth donor and the known dose added at the laboratory.

opposite the source, which is typical for dental x-ray examinations) and non-zero readouts in nonirradiated parts of the teeth. The findings (table 3) are not statistically significant and are given only as guidance to demonstrate general tendencies. Unfortunately, x-ray doses for all the teeth were below the threshold of reliable dose reconstruction with the EPR technique, and therefore these figures could not be used to correct the results of the intercalibration.

Table 3. Dose assessments for different parts of teeth (SCRM) (in mGy)

Tooth number	Inner part (unexposed)	External part (unexposed)	Inner part (exposed)	External part (exposed)	Mean dose of exposed half
T4	30	50	230	240	230±50
T5	30	50	230	270	250±50
T6	20	50	290	300	300±70

UU measurements were taken at a microwave power of 2 mW (a method chosen after a series of tests as the most promising one to minimize noise and, therefore, uncertainty of the dose determination). An analysis of expected uncertainties was performed before analysis, and the number of spectra to be collected at each power was set at 42 for the irradiated samples, 12 at the additive dose level of 1 Gy, and 6 at the 10-Gy additive dose level. Samples were stored for a minimum of 12 hours at room temperature following each irradiation in a ^{60}Co irradiator at a dose rate of 10 Gy/h (5 mm of aluminum was used for electron buildup). Dose increments were 210, 435, 435, 435, 870 mGy.

Both sets of dose determinations are presented in table 4. As expected, the results of the second stage intercalibration were not as clear-cut as those of stage 1, and they could not be interpreted definitely. Both laboratories demonstrated good agreement (within 17%) with nominal

Table 4. Intercomparison of whole teeth exposed *in vitro*

Group	Sample	Laboratory-added dose mGy	Laboratory	Measured dose mGy
1	T1	171	UU	190±50
	T4		SCRM	230±50
2	T2	256	UU	180±50
	T5		SCRM	250±50
3	T3	200	UU	190±100
	T6		SCRM	300±70

dose values, although the results produced by the SCRM technique tended to overestimate the doses. The latter may be explained by the contribution of the lifetime dose (particularly medical x-ray exposure) to the total dose to the tooth, which was determined by the dose reconstruction. Neither data on x-ray examination nor age of patients was available, making assessment of this component of the total dose impossible. Since the detected doses corresponding to the pre-intercalibration history of the teeth were found to be below the threshold of reliable dose reconstruction, the correction of the results was, unfortunately, impossible.

Stage 3. Intercomparison of Teeth From Liquidators

The third stage of cross-calibration was designed to test the capability of the two techniques to perform dose reconstruction using teeth exposed *in vivo*. Since the actual doses were unknown and, therefore, absolute validation of the results is impossible, the expected yield of this effort was a consistency check.

Intercomparison Design

The initial intention was to provide both laboratories with identical samples exposed *in vivo*. Three groups of samples from liquidators totaling 34 specimens were shipped to the United States:

- 13 halves of large teeth (molars)—the remainders were retained at SCRM
- 15 teeth from pairs extracted simultaneously from the same individual and therefore presumably having the same doses
- 6 samples in the form of pieces of mechanically separated tooth enamel

Due to the limited time that could be allocated by UU for examination of the samples, the number of dose reconstructions was reduced to five. The specimens selected (numbers X23, X24, X25, X26, and X28) were represented by granular samples of tooth enamel. For each sample, the initial separation of tooth enamel was performed at SCRM using a steel dental drill. After the removal of dentine, the pieces of enamel were collected and the whole sample was divided into parts of about 100 mg each for independent determination of dose by the participants. The characteristic size of enamel grains was about 500 micrometers, although the dimensions of individual particles varied from hundreds of microns to several millimeters.

Methods and Results

SCRM used basically the same standard technique described above. The technique used at UU was somewhat modified. The parameters used for the second EPR intercomparison of the liquidators' teeth were as follows: 8-mW microwave power, 5-Gauss modulation amplitude, 20-second conversion time, 35-Gauss sweep width, 168-ms time constant, and 10^5 gain.

The dose reconstruction was done using the spectra from the samples, a baseline enamel sample with negligible dose, and an empty EPR tube. The spectrum of the tube was subtracted from each spectrum taken of all the samples and the baseline samples. This was done in proportion to the number of scans taken in each enamel spectrum, that is, an enamel spectrum composed of 12 sweeps was corrected by subtracting the spectrum of the empty tube, which itself was normalized to 12 sweeps. The spectra of the samples were then precisely normalized to the standard of 10 sweeps and 100 mg per spectrum by the normalization factor:

$$(10 \text{ sweeps}/\#\text{ of sweeps taken}) \bullet (100 \text{ mg sample weight})$$

This adjustment was not necessary for the baseline sample as it was precisely weighed before measurement. The spectrum of the baseline sample was then subtracted from all the spectra of the irradiated and unirradiated enamel samples. From these resulting background and baseline-free spectra, we did the dose reconstruction on the g-perpendicular signal extremes using standard least squares fitting and error propagation for the dose estimates and errors, respectively. The additive dose technique was employed using only one applied dose of 5 or 10 Gy (10 Gy if the sample mass was less than 35 mg). The number of spectra taken at each dose was 20 to 25 for the zero dose and 12 for the one applied dose (25 if the sample mass was less than 35 mg).

In this intercomparison, an additional check of the purity of the samples was performed using the EPR spectra recorded before additional irradiation and the spectrum of a milk tooth as the standard of the background signal. Two (X24 and X25) of five samples had satisfactory purity. Three others were subjected to treatment with an NaOH solution. After the chemical treatment, the shape of the signal had improved. Two of the samples (X26 and X28) were considered to be purified completely. Although the purity of the third one (X23) had improved, it still had some distortions in the spectrum. Because of significant loss of mass (it had decreased from 70 to 33 mg), we decided to refrain from further purification and proceed with EPR measurements.

This third stage of cross-calibration had relatively poor results (table 5). The doses, determined in different laboratories, coincided within declared uncertainty ranges for only two individuals out of five. The results from the two laboratories differed significantly (up to 60%) for some of the samples. The results of this intercomparison are discussed elsewhere [6].

At the present stage of the intercomparison, it is impossible to determine the major reasons for these discrepancies. Adequate interpretation of the results requires additional investigation and, possibly, conducting the intercomparison with a partially modified design. One possibility could be to use teeth from individuals whose doses have been assessed by independent methods of retrospective dosimetry (such as a FISH test or analytical dose reconstruction).

Table 5. Results of intercomparison with teeth of liquidators exposed *in vivo*

Sample	SCRM, Gy	UU, Gy
X23	0.36±0.05	0.67±0.10
X24	1.42±0.14	1.60±0.24
X25	1.08±0.11	1.56±0.23
X26	1.50±0.15	1.56±0.23
X28	0.48±0.05	1.18±0.18

Discussion

The cross-calibration performed within Task 2 of the project was the first international, full-scale effort to harmonize EPR-dosimetric techniques developed in Ukrainian and US laboratories as well as to perform quality assurance of this method. The three stages of cross-calibration, for the most part, covered all degrees of complexity and adequacy of approaches to dose reconstruction. Generally positive, the results of the cross-calibration have proven the applicability of EPR dosimetry to practical reconstruction of individual doses.

The reproducibility of the results of the different versions of EPR technique that were designed and used on different continents is, at worst, within the 60% standard deviation interval. Clearly, even with this conservative and potentially improvable uncertainty interval, EPR dosimetry could produce more accurate dose assessments than any other method of retrospective dose reconstruction to be used in a post-Chernobyl epidemiological followup. Moreover, stage 2 of the cross-calibration experimentally demonstrated that lifetime diagnostic x-ray examinations may lead to overestimation of doses within only

30% limits. This important point, which needs additional investigation, could resolve positively the greatest concern presently associated with the use of EPR dosimetry for reconstruction of individual doses among the liquidators.

Task 3
Test of Practical Dose Determination

A system for retrospective reconstruction of doses received by the Chernobyl liquidators was tested in Task 3.

Dose Reconstruction

The teeth from liquidators were collected in the course of dental surgical practice in the Kiev central liquidators' polyclinics. Extracted teeth were accompanied by special ID forms (see appendix) reflecting the personal data necessary for tracing the individual, information about occupational contacts with ionizing radiation, lifetime medical x-ray examination of the head, data on location of the extracted tooth, and the diagnosis leading to extraction. After extraction, the teeth were preserved in formalin in small bottles. Periodically (approximately once a month), the teeth were transported to the Laboratory of External Exposure Dosimetry for storage, processing, and determination of radiation doses.

Upon arrival, all teeth were subjected to preprocessing, including rinsing in distilled water and drying at 80 °C. The tooth root was separated and, if necessary, residuals of soft tissues and damaged areas of teeth were removed. Then, teeth were placed in intermediate storage under room conditions.

The dose determination cycle began with chemical treatment as discussed in Task 1 and illustrated in figure 2. The samples of pure tooth enamel were subjected to measurements, including recording of EPR-spectra (with parameters as indicated in table 1) and laboratory irradiation with preset doses. Individual calibration curves were plotted for all measured samples, and doses were determined accounting for individual radiosensitivity of enamel. It was found that calibration

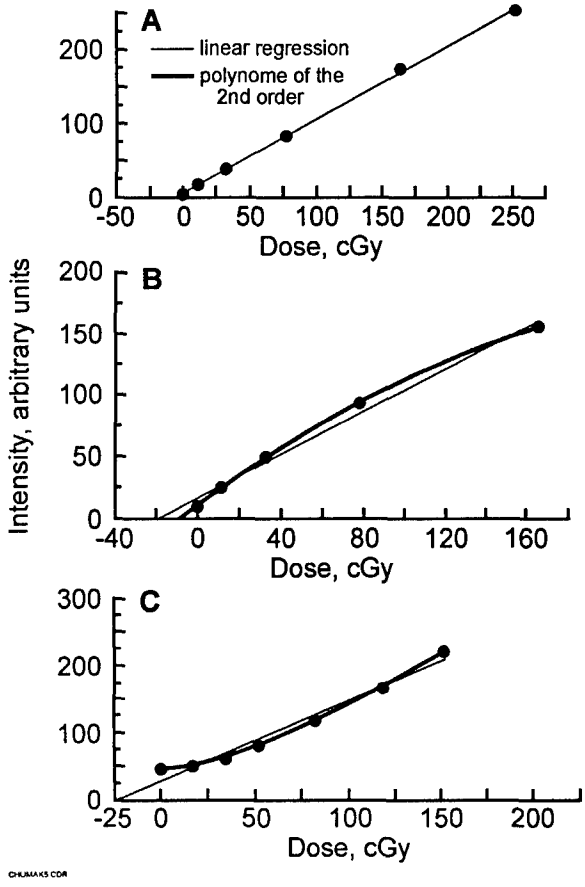

Figure 5. Different types of dose-response (calibration) curves for tooth enamel samples. (A) linear dose response (95% of samples), (B) superlinear dose response (4% of samples), (C) sublinear dose response (1% of samples).

curves may not always be fitted by linear regression. In some cases, dose-response curves were superlinear (figure 5b) or sublinear (figure 5c). Although the total fraction of teeth with nonlinear dose-response curves was rather small (about 5%), this phenomenon needs to be accounted for in order to avoid significant under- or overestimation of dose values. The sources of this effect need to be studied and localized.

The results of dose reconstruction of 146 teeth from 135 liquidators are shown in table 6. The cumulative doses and dose values are not corrected for lifetime exposure. Age of the patient and type of tooth give a clue to the amount of the dose due to natural background. The type of tooth is also important for the possible contribution of ultraviolet (UV) light to the generation of paramagnetic centers, which may be more pronounced for the front teeth (classes A and B).

The frequency distribution of individual doses measured with EPR dosimetry is presented in figure 6. It may be seen that the shape of the distribution is close to lognormal—mean dose is 287 mGy, geometric mean is 205 mGy. Median dose value is 200 mGy. The reconstructed dose values range from 30 to 2220 mGy. The individual with the highest dose is a policeman who performed his guard mission outdoors during the first days after the accident. On some occasions, several teeth used in the investigation came from the same individual.

Table 6. Individual dose values reconstructed in course of EPR dosimetric exercise

Sample number/ ID code	Age at time of tooth extraction	X-ray examinations	Beginning of cleanup work (year)	Type of tooth*	Total dose (cGy)
1	2	3	4	5	6
177/13862	60	–	86	A	39
178/16042	53	–	86	C	16
180/17568	25	–	86	C	5.5
182/2069	56	+	86	C	13
183/59	45	–	86	C	24
185/10709	43	+	86	C	12
187/15961	61	–	86	C	19
190/17871	64	–	86	C	14
				C	29
192/17099	60	–	86	C	25
194/20457	45	–	86	C	10
195/13337	58	–	86	A	34
198/12006	55	+	86	B	15
199/20491	44	+	86	C	11
200/7107	46	–	86	C	10
201/4385	43	–	86	C	17
202/1304	56	+	86	C	28
204/1801	47	+	86	C	8
205/	40	+	86	C	13
208/9731	56	–	86	C	12
301/15518	60	+	86	C	19
302/16012	58	–	86	C	28
303/13827	39		86	U	6
304/13473	55	+	86	C	15
305/7826	39		86	A	31
307/4513	48	–	86	C	8
278/14877	40	–	86	C	4
28/10058	55	+	86	C	12
281/14571	57	–	86	C	11
284/17631	57	–	86	A	31
				A	16
287/19245	57	–	86	C	18
298/8822	66	–	86	B	34
33/483	57	–	86	A	46
8/4416	44	+	86	U	18
				A	21
4/13930	53	+	86	C	45
				C	67
39/15579	62	+	86	B	78
16/4689	52	+	86	C	38
9/7821	55	–	86	C	53

*A – incisors and canines, B – premolars, C – molars, U – unknown

Table 6. Continued

1	2	3	4	5	6
10/11338	53	+	86	A	40
15/13919	59	–	86	C	22
29/16047	53	+	86	C	19
24/6902	55	–	86	A	64
53/1926	54	+	86	C	18
46/7125	46	–	87	C	13
97/10075	56	–	86	A	55
98/199	52	–	86	A	59
43/17164	55	+	87	C	120
67/16344	38	+	86	A	66
138/17320	41	+	87	C	9
151/3598	64	+	86	C	12
132/3915	66	–	86	A	23
64/3978	40	+	86	C	27
84/2743	54	–	87	C	20
181/9287	63	+	86	B	14
6C	53	–	86	C	142
19C	62	–	86	C	25
105/11735	44	–	86	C	16
106/9138	44	–	86	C	20
108/573	62	+	86	C	30
				C	23
7/1670	62	+	86	A	30
71/763	64	–	86	C	18
72/18249	42	–	86	C	3.5
78/18414	30	+	86	C	5
				C	8
79/15694	79	–	86	C	13
82/7452	49	–	86	C	5
83/18187	49	+	86	C	13
86/17174	50	–	87	C	12
87/15171	55	–	86	C	6
35/3602	28	+	86	C	20
41/4068	37	+	86	C	18
42/3304	55	–	86	C	7
44/8329	47	–	86	B	18
61/8329				B	28
45/8203	60	+	86	B	20
49/9037	65	+	86	C	19
56/9737	56	–	86	A	29
59/7727	45	–	86	C	8
50/7727				C	6
65/4299	50	+	86	C	9
68/10373	50	+	87	C	8

*A – incisors and canines, B – premolars, C – molars

Table 6. Continued

1	2	3	4	5	6
69/17092	60	-	86	C	8
109/17545	45	-	87	C	21
113/8025	29	-	87	C	6
123/13820	52	+	86	A	50
125/10396	55	+	86	C	14
129/13494	57	+	86	A	24
130/15240	64	-	86	A	60
142/5009	68	-	86	A	71
145/4768	56	-	86	A	35
148/16267	49	+	87	C	67
153/13349	60	+	86	C	96
209/1408	40	-	86	U	47
212/8456	46	-	86	A	39
216/8544	45	-	86	C	25
217/13506	43	-		C	13
218/13953	59		86	U	20
219/15067	64	+	86	U	13
223/3467	57	-	86	C	23
224/18355	71	-	87	C	65
225/17113	56	-	87	C	14
227/18318	55	-	86	C	10
228/8369	53	+	86	C	14
249/8369	54			C	39
230/2721	57	-	86	C	12
231/13381	63	-	86	C	195
234/16478	63	-	86	C	16
235/8152	56	+	86	C	23
236/4325	47	-	86	A	27
238/9123	62	-	86	A	70
247/14939	43	-	86	C	18
250/6863	55	+	86	C	21
				C	15
251/7583	45	+	86	A	23
263/8939	64	-	86	A	35
300/17933	66	+	86	C	40
81/10298	33	-	89	C	8
308/19933	51	-	87	C	7
309/8120	56	-	88	C	13
312/2043	61	+	86	C	23
313/16137	68	-	87	C	26
314/4571	58	+	86	U	64
317/21873	55	-	86	C	50
320/7004	48		86	C	6
321/20577	44	-	87	C	7

*A – incisors and canines, B – premolars, C – molars, U – unknown

Table 6. Continued

1	2	3	4	5	6
322/15777	38	-	86	A	36
				B	31
325/23887	42	+	86	C	8
326/15707	64	-	86	A	42
330/17012	61	+	86	C	12
331/8719	33	+	86	C	10
332/14533	59	-	86	C	35
334/3713	57	+	86	C	12
335/15336	51	-	86	B	3
337/4722	68	-	86	A	21
340/13695	52	-	86	C	32
				C	20
2C			86	U	57
17C	40		86	U	30
18C	35		86	U	222
20C	41		86	U	40
21C			86	U	30
22C	37		86	U	15

*A – incisors and canines, B – premolars, C – molars, U – unknown

The doses generally depend on the amount of time spent working in the 30-km zone. As may be seen from table 6, most of the liquidators involved in the current dose reconstruction effort began their work in 1986. Median dose of this group is 211 mGy, while doses to liquidators of 1987 and later years are lower—164 mGy. Maximum doses to liquidators from 1986 and 1987 were 2220 mGy and 1200 mGy, respectively. This observation is in good agreement with the fact that the most dose-intensive activities were performed during the first months after

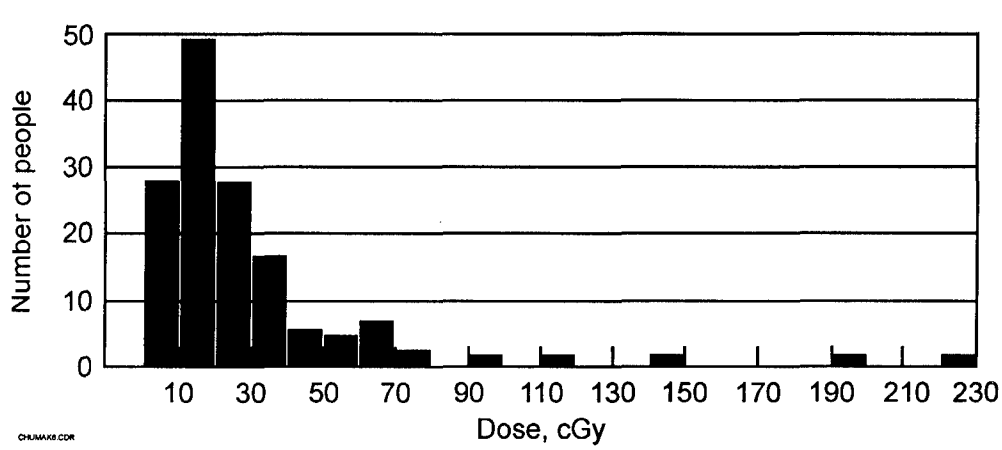

Figure 6. Frequency distribution of individual doses to liquidators determined by EPR dosimetry of teeth.

the accident, when dose rate levels were much higher and most of the cleanup work took place.

System Development

Practical demand dictates a need for reconstruction of radiation doses to the large groups of liquidators included in cohorts studied for epidemiological followup. The possibility of long-term storage of tooth samples together with increasing performance of the EPR-dosimetric technique make this task feasible.

However, since the teeth used for dose reconstruction are extracted for medical reasons only, sampling is a random and relatively infrequent event. Besides, the process of natural tooth loss is an important factor, reducing the available sampling population over time.

Therefore, a systematic approach to dose reconstruction from teeth, including sample acquisition, is required. For longitudinal epidemiological followup of an exposed population, the problem of availability of samples may be solved by organizing a widespread network for acquisition of teeth extracted from the members of the studied cohort. This network should be based on centers with a high density of liquidators and other heavily exposed populations. Since the productivity of EPR dosimetry is limited and not yet sufficient to process all the potential influx of samples in real time, a central bank of bioprobes should be established for acquisition, storage, processing, and retrieval of tooth material. Potentially, every participant of this studied cohort sooner or later would be covered by this effort, yielding tooth samples to the bioprobe bank. Teeth from those individuals who were included in the study cohort and have died could be received in the course of autopsy. Dose values, reconstructed by means of EPR, could be entered in the personal dosimetric file of the individual for access by radioepidemiologists.

Development of such an infrastructure for dose reconstruction is underway now in Ukraine. The acquisition network (figure 7) would be based on special liquidators' hospitals in seven regional centers, covering about 45% of the heavily exposed cleanup workers. The samples, along with ID forms, would be transferred to the central bioprobe bank for long-term storage and subsequent processing.

The role of the central bioprobe bank is to coordinate activities in acquisition of teeth, EPR dosimetry, and data management on a national scale. Results of the ongoing EPR dose reconstruction will be forwarded to the National Registry and sent in parallel to the local health care bodies in order to provide feedback to individuals whose teeth had been submitted for examination. Access to the individual

dose records will also be provided to the researchers involved in post-Chernobyl followup studies.

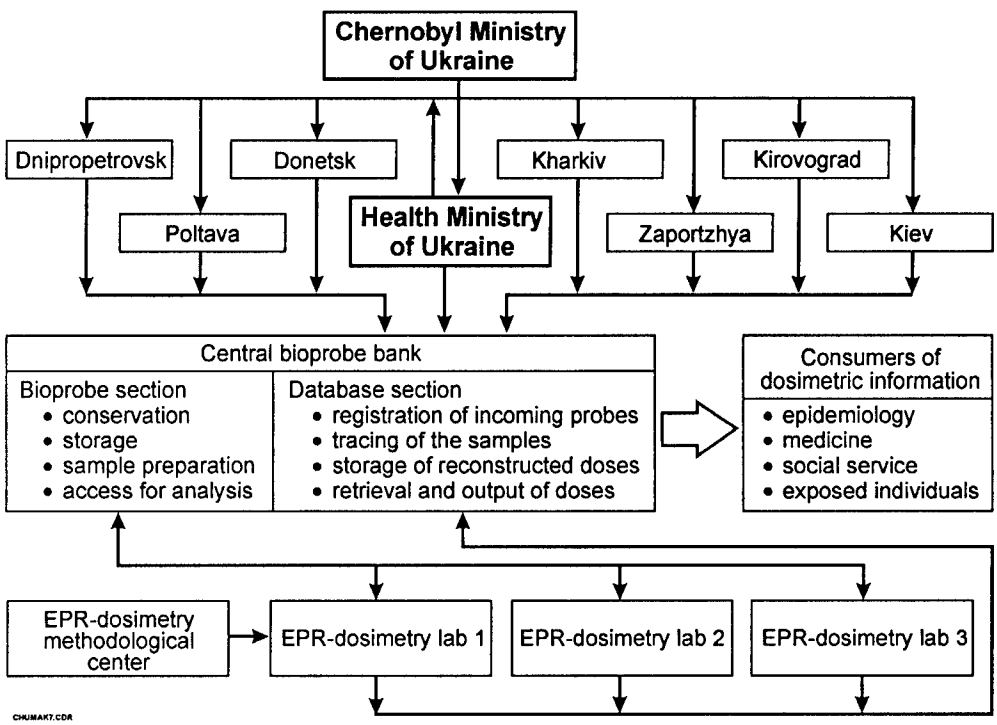

Figure 7. Infrastructure of the system for retrospective reconstruction of doses received by the Chernobyl liquidators

Discussion

Although performance and capability were tested in a series of cross-calibrations and routine dose reconstructions, several issues of key importance need to be resolved prior to extensive use of EPR dosimetry with teeth for followup studies. These yet unresolved problems may threaten the utility of EPR dosimetry. Some of these effects have been known for a long time; others were discovered recently. Among these are the well-known effect of enhanced sensitivity to low-energy photons and the recently reported generation of paramagnetic centers by UV light [7]. Nonlinearity of dose-response curves in the dose range below 1 Gy was observed by the authors of this report only during the dose reconstruction exercise and is yet unpublished.

Effects of Medical X Rays

Irradiation of tooth enamel with low-energy photons may lead to substantial (up to seven times) overestimation of the tissue-absorbed dose. This effect has pronounced energy dependence, with the highest oversensitivity at 60 keV. The signals from paramagnetic centers produced by high-energy (accidental) and low-energy (medical x-ray) photons are identical, making discrimination of these signals by means of EPR impossible. As a result, a dose measured by EPR is the sum of an accidental component (dose of interest) and a component due to medical exposure. The degree of significance of the latter depends on the relative value, which is a function of incidence energy, dose per examination, and number of examinations. This means that the type of x-ray apparatus used in the dental practice is very important, determining, after all, the degree of significance of the x-ray component.

In order to clarify this issue, it is necessary to conduct a systematic investigation of the effects connected with x-ray exposure. This inves-

tigation should include both experimental and theoretical evaluation of dose responses prompted by different types of x-ray examination, including different geometry, x-ray apparatus, and dose per examination. The issue of doses deposited on neighboring and opposite teeth should be studied also. The contribution to the total tooth dose from different types of x-ray examination (e.g., gamma-tomography, panoramic diagnostics of the mandible, skull and sinus films) and radiotherapy is still unknown and requires special investigation. This work will demand the use of both mathematical and physical phantoms for the simulation of realistic situations.

Since x-ray practices are different in Ukraine and the United States, special attention in this research should be paid to comparison of these two cases and the development of approaches to the solution of this problem. Intercomparisons using teeth exposed to x rays or exposed to mixed fields would be useful as well. Such studies should either conclude that the x-ray contribution to the tooth dose is insignificant (and establish the limits of the application of this assumption) or else recommend how to mitigate or account for the effect of x-ray examination.

Analysis of different teeth from the same person may be useful for understanding of the effects of x-ray examination on different types of teeth and other factors. Among the 135 individuals studied in the present research, multiple teeth were available from 11 persons. In nine cases, teeth were extracted simultaneously, with presumably equal accidental doses. In two cases (individuals 190/17871 and 284/17631), the doses determined using different teeth showed a discrepancy above the 40% standard deviation accuracy granted by the technique. Since the teeth in both cases were of the same type, and x-ray examination was not reported on the ID form, this phenomenon could not be explained by the contribution of x-ray exposure or variations in the type of tooth. Some as yet unknown effects may be responsible for such deviation. Unfortunately, the limited scope of research and similarities in lifetime exposure and type of teeth give little material for analysis. However, so far, the largest deviation of doses determined for similar teeth was 52%, which is not a particularly large error, considering errors typical for other methods of retrospective dosimetry.

Effects of UV Light

Another phenomenon which may affect the reliability of dose reconstruction with tooth enamel is the generation of paramagnetic centers by UV light. Information about the role and the qualitative and quantitative characteristics of this effect is quite contradictory. This effect was first discovered and reported by Ivannikov et al. [7] in 1995. The

series of experiments conducted worldwide to study this effect brought no clarification.

According to existing information and our own data, the centers generated in tooth enamel have position and shape very similar to those of radiation-induced centers. That means that discrimination of UV- and radiation-induced signals by spectrometric means may be difficult. It is expected that UV irradiation effects are most pronounced for front teeth; such factors as time spent outdoors and elevation of the living area above sea level may also influence the degree of this effect.

At present, the problem of UV irradiation needs to be approached in a systematic way; this phenomenon must be studied from the point of view of its physical, spectrometric, and kinetic (half-life) properties. Processes of generation of paramagnetic centers as a function of wavelength and intensity of UV light and decay of these centers should be investigated in order to obtain a clear view of this effect.

Study of spectrometric properties (e.g., saturation of the signals) may yield an approach to discrimination of UV- and radiation-induced signals by means of EPR technique. Investigation of depth profiles of UV-generated signals in teeth for different energies of UV photons and the UV component of daylight should help explain attenuation of UV light in enamel and could be used for target etching of exposed fractions of tooth enamel. Recommendations concerning accounting for and mitigating this effect should be issued as a final point of this research.

Nonlinearity of Dose Response Curves

Nonlinearity of dose response curves in the dose range below 1 Gy was observed in some teeth in the course of the dose reconstruction exercise in this project. Before, saturation of the dose-response curve was observed only at doses above 10 Gy; below this range, the curve was considered to be linear, and this property is widely used for extrapolation of calibration curves in the low-dose regions. Moreover, the techniques based on the utility of a single calibration factor (without additive dose) critically depend on linearity of the dose-response function.

Nonlinearity of dose response curves may have a significant influence on the results of dose reconstruction. Not accounting for nonlinearity of calibration curves leads to substantial under- or overestimation of individual doses (as illustrated in figure 5). Advanced study of this effect, investigation of factors having impact on the dose-response curve, and the development of methods for extrapolating additive-dose curves are necessary for accurate and reliable retrospective dosimetry

using teeth as a natural dosimeter. Since this effect takes place in only about 5% of cases, the scope of dose reconstruction should be large enough to provide consistent and statistically significant conclusions.

Summary

During the 14-month period covered by this contract, extensive research and technological developments were performed at SCRM AMS Ukraine in close collaboration with the University of Utah, USA. As a result of this effort, EPR dosimetry with teeth was brought to the level of a semiroutine technique for evaluation of doses received by individuals heavily exposed after the Chernobyl accident.

Special attention was paid to quality assurance for this high-technology method in order to provide accurate and reliable individual dose assessments. The quality assurance program included several international cross-calibrations using a variety of specimens, from pulverized tooth enamel in the beginning to whole teeth from liquidators exposed *in vivo* during the final phase of intercomparison.

Since the limited availability of samples from the individuals of interest is one of the important bottlenecks of EPR dosimetry now, a complete system for the reconstruction of doses to liquidators must include a means for acquiring the samples. Therefore, an organization pattern for acquisition of teeth extracted by medical prescription from the Chernobyl liquidators was presented. This infrastructure is being implemented in Ukraine now.

The semiroutine technique developed and adopted over the period of consideration was used for retrospective dosimetry of a considerable group of liquidators. In total, doses were reconstructed for 135 individuals who took part in the Chernobyl cleanup in 1986–87. The cohort of liquidators studied was assembled randomly in the course of dental surgery in the Kiev central liquidators' polyclinic.

Analysis of the data obtained revealed that the mean dose of this group is 287 mGy, ranging to the highest value of 2220 mGy. This is signifi-

cantly higher than the officially reported mean dose of 110 mGy. Therefore, the widely accepted opinion that the official records are of low quality and underestimate the actual doses was supported by this first retrospective dosimetry effort involving an appreciable number of subjects. The fact that doses reconstructed instrumentally are much higher than those officially recorded gives additional justification for the investment in development and performance of retrospective dosimetry, particularly EPR.

However, recent research and developments in the field of EPR dosimetry make obvious a need for further investigations. From the pragmatic point of view, these investigations should be conducted along the following lines:

- Investigation and development of approaches to account for EPR signals induced by lifetime medical x-ray exposure
- Comprehensive study of the effects in tooth enamel caused by UV light
- Investigation of the factors causing nonlinearity of the dose-response function in the dose range below 1 Gy and development of approaches to account for this effect in dose determination
- Cross-validation of EPR dosimetry with independent methods of retrospective dosimetry; this may be achieved by parallel application of different methods (e.g., EPR, FISH, and analytical) to the same objects
- Methodological research aimed at improving the technological capabilities of EPR dosimetry and enhancing the productivity of the technique.

Completion and success of the outlined efforts will bring EPR dosimetry from a quite exotic methodology to an ordinary dosimetric routine like gamma-spectroscopy and alpha counting.

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Appendix

Identification Form for Tooth Sampling

1. Complete affiliation of the hospital which performed extraction

2. ID number _____ 3. Date of extraction ___/___/___

N	General information	Fragment 1
1	Family name	
2	First name	
3	Second name	
4	Sex (male - 1, female - 2)	
5	Date of birth	
6	Liquidators pass (series and number)	
7	Year of work in Chernobyl	
8	Dose value, officially recorded (if available)	
9	Date of evacuation from the 30-km zone	
10	From what settlement	

N	Postal address at present time	Fragment 2
1	ZIP code	
2	Region	
3	District	
4	Town	
5	Street	
6	House	
7	Building	
8	Appartment	

4. Places of stay since the accident (region, district, settlement)
(1986 in all details, afterwards - reflect locations with period of stay more than 3 months).

Year	Settlement	Period of stay	
		Arrival	Departure

5. Professional contact with radiation (including military service) _____

_____

6. Information about x-ray examinations of skull, jaws, teeth (dates, type, approximate number during life span): _____

_____

7. General deseases affecting solid tissues of tooth _____

_____

_____

_____

_____

8. Location of the tooth and reason of extraction:

8	7	6	5	4	3	2	1	1	2	3	4	5	6	7	8

9. Affiliation during the Chernobyl recovery activities

10. Notes

11. Name of physician who extracted the tooth

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13. ABSTRACT *(Maximum 200 words)*

Accurate, reliable dose reconstruction is a critical component in the epidemiological followup of liquidators. Dosimetry of teeth by electron paramagnetic resonance (EPR) is a state-of-the-art laboratory technique that is key to this effort. The Scientific Center of Radiation Medicine (SCRM) has developed and refined this technique in order to meet the practical demands of large-scale epidemiologic followup of the Chernobyl liquidators. Independent analysis using similar technology was performed by investigators at the University of Utah and showed good correlation with the SCRM results. The lower limit of detection for reliable dose reconstruction was 100 mGy. Techniques were applied to samples from approximately 135 liquidators involved in cleanup activities within the first 2 years after the Chernobyl accident in 1986. Mean dose was 287 mGy, geometric mean was 205 mGy, and median dose value was 200 mGy. The reconstructed dose values range from 30 to 2220 mGy. Correlation of results between the two institutions was generally within 17%. This report also addresses some confounding factors (previous medical x-ray exposures, ultraviolet light effects on anterior teeth, nonlinearity of dose response curves below 100 mGy) and how to deal with them.

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